



US006440420B1

(12) **United States Patent**
Liu et al.

(10) Patent No.: **US 6,440,420 B1**
(45) Date of Patent: **Aug. 27, 2002**

(54) **METHOD FOR EXTRACTING OLEAGINOUS SUBSTANCES FROM GERMINATION-ACTIVATED *GANODERMA LUCIDUM* SPORES**

(76) Inventors: **Xin Liu; Xiao-Ni Huang**, both of Building No. 391, 135 Xingang Xi Road, Guangzhou (CN); **Peter Chee-Keung Chung**, Room 1505, Argyle Centre, 688 Nathan Road, Mongkok, Kowloon (HK)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

(21) Appl. No.: **09/810,213**

(22) Filed: **Mar. 19, 2001**

(51) Int. Cl.⁷ **A61K 35/78**

(52) U.S. Cl. **424/195.15; 435/254.1**

(58) Field of Search **424/195.15; 435/254.1**

(56) **References Cited**

U.S. PATENT DOCUMENTS

4,675,198 A 6/1987 Sevenants
5,017,397 A 5/1991 Nguyen et al.
6,111,108 A 8/2000 Lopez-Avila et al.

OTHER PUBLICATIONS

Wasser, Solomon P. et al.; Therapeutic Effects of Substances Occurring in Higher Basidiomycetes Mushrooms: A Modern Perspective; Critical Reviews in Immunology, 1999, vol. 19, P 65-96.

Lin, Lee-Juan et al.; Separation of oxygenated triterpenoids from *Ganoderma lucidum* by high-performance liquid chromatography; Journal of Chromatography, 1987, vol. 410, P 195-200.

El-Mekkawy, Sahar et al.; Anti-HIV-1 and Anti-HIV-1-Protease Substances from *Ganoderma lucidum*; Phytochemistry, 1998, vol. 49, No. 6, P 1651-1657.
O'Neil, Carol E. et al.; Basidiospore Extracts: Evidence for Common Antigenic/Allergenic Determinants; Int. Archs Allergy appl. Immun., 1988, vol. 85, P 161-166.

Kim, Kug Chan et al.; *Ganoderma lucidum* extract protects DNA from strand breakage caused by hydroxyl radical and UV irradiation; International Journal of Molecular Medicine, 1999, vol. 4, P 273-277.

Min, Byung-Sun et al.; Triterpenes from the Spores of *Ganoderma lucidum* and Their Inhibitory Activity against HIV-1 Protease; Chem Pharm Bull, 1998, vol. 46(10), P 1607-1612

Kino et al.; An immunomodulating protein, Ling Zhi-8 (LZ-8) prevents insulinitis in non-obese diabetic mice; Diabetologia, 1990, vol. 33, P 713-718.

Gengtao, Liu et al.; Some Pharmacological Actions of the Spores of *Ganoderma lucidum* and the Mycelium of *Ganoderma Capense* (Lloyd) Teng Cultivated by Submerged Fermentation; Chinese Medical Journal, 1979, vol. 92(7), P 496-500.

Primary Examiner—Francisco Prats

Assistant Examiner—Susan D. Coe

(74) Attorney, Agent, or Firm—Fei-Fei Chao; Venable, Baetjer, Howard & Civiletti, LLP

(57) **ABSTRACT**

The present invention relates to a method for extracting the oleaginous substances from sporoderm-broken *Ganoderma* spores using SCF—CO₂. The method contains the steps of: (1) inducing germination of *Ganoderma* spores by incubating the spores in a nutritional solution; (2) activating the *Ganoderma* spores by placing the germination-induced spores in a well ventilated culture box kept at constant temperature and humidity; (3) breaking the *Ganoderma* spores by a mechanical means to obtain the sporoderm-broken spores; and (4) extracting the oleaginous substances from the sporoderm-broken spores using a supercritical fluid—carbon dioxide (SCF—CO₂) extraction method. The preferred supercritical conditions include 5 M to 60 M Pa of pressure; 32° C. to 85° C. of temperature; and 5 kg/h to 80 kg/h of flow capacity rate. The total extraction time in SCF—CO₂ is between 0.5 hour to 6 hour. The method produces approximately 37% by weight of oleaginous substances from the sporoderm-broken *Ganoderma* spores. These oleaginous substances are transparent and contain the special fragrance of *Ganoderma* spores. There is no trace of deposit, solvent residue, or oxidization in the oleaginous substances.

19 Claims, No Drawings